

SEQUENCE LISTING

<110> Novozymes A/S

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Andersen, Carsten

Borchert, Torben Vedel

<120> Pullulanase variants and methods for preparing such variants with predetermined properties

<130> 6072.204-US

<140> To Be Assigned

<141> 2001-08-21

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<170> PatentIn version 3.1

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Phe Ser Ser Met Ile His Leu Arg Asn Gln His Pro Ala Phe Arg Met	820	825	830	
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 Glu Val Ile Tyr Glu Ala His Val Arg Asp Phe Ser Ile Asp Ala Asn
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 Ser Gly Met Lys Asn Lys Gly Lys Tyr Leu Ala Phe Thr Glu His Gly
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atc tat gaa atg gat gtc cgt gac ttt tcc att gac cct aat tcg ggt Ile Tyr Glu Met Asp Val Arg Asp Phe Ser Ile Asp Pro Asn Ser Gly 435 440 445	1344
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ggc cct gac aac gta aag acg ggg ata gat tcc tta aaa caa ctt ggg Gly Pro Asp Asn Val Lys Thr Gly Ile Asp Ser Leu Lys Gln Leu Gly 465 470 475 480	1440
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aca gtg gcc tat gaa tta act gat cat gtt aat aaa gac aaa tgg gga Thr Val Ala Tyr Glu Leu Thr Asp His Val Asn Lys Asp Lys Trp Gly 850 855 860	2592
aat atc att gtt gtt tat aac cca aat aaa act gta gca acc atc aat Asn Ile Ile Val Val Tyr Asn Pro Asn Lys Thr Val Ala Thr Ile Asn 865 870 875 880	2640
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Ser Ala Asp Ile Pro Gly Asn Pro Ser Gln Val Gly Ile Ile Val Arg 50 55 60
Thr Gln Asp Trp Thr Lys Asp Val Ser Ala Asp Arg Tyr Ile Asp Leu 65 70 75 80
Ser Lys Gly Asn Glu Val Trp Leu Val Glu Gly Asn Ser Gln Ile Phe 85 90 95
Tyr Asn Glu Lys Asp Ala Glu Asp Ala Ala Lys Pro Ala Val Ser Asn 100 105 110
Ala Tyr Leu Asp Ala Ser Asn Gln Val Leu Val Lys Leu Ser Gln Pro 113

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Ala	Asn	Lys	Asp	Ile	Pro	Val	Thr	Ser	Val	Lys	Asp	Ala	Ser	Leu	Gly
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Gln	Asp	Val	Thr	Ala	Val	Leu	Ala	Gly	Thr	Phe	Gln	His	Ile	Phe	Gly
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Phe	Ser	Tyr	Ile	Pro	Ser	Thr	His	Ala	Val	Tyr	Asp	Thr	Ile	Asn	Asn
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Pro	Asn	Ala	Asp	Leu	Gln	Val	Glu	Ser	Gly	Val	Lys	Thr	Asp	Leu	Val
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Thr	Val	Thr	Leu	Gly	Glu	Asp	Pro	Asp	Val	Ser	His	Thr	Leu	Ser	Ile
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Gln	Thr	Asp	Gly	Tyr	Gln	Ala	Lys	Gln	Val	Ile	Pro	Arg	Asn	Val	Leu
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305					310					315					320
Thr	Gln	Lys	Ala	Thr	Thr	Phe	Lys	Val	Trp	Ala	Pro	Thr	Ser	Thr	Gln
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Val	Pro	Met	Thr	Ala	Ser	Gly	His	Gly	Val	Trp	Glu	Ala	Thr	Val	Asn
		355					360					365			
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370

375

380

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Gly Thr Arg Gly Met Ile Val Asp Leu Ala Lys Thr Asp Pro Ala Gly
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Trp Asn Ser Asp Lys His Ile Thr Pro Lys Asn Ile Glu Asp Glu Val
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Ile Tyr Glu Met Asp Val Arg Asp Phe Ser Ile Asp Pro Asn Ser Gly
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Met Lys Asn Lys Gly Lys Tyr Leu Ala Leu Thr Glu Lys Gly Thr Lys
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Gly Pro Asp Asn Val Lys Thr Gly Ile Asp Ser Leu Lys Gln Leu Gly
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Ile Thr His Val Gln Leu Met Pro Val Phe Ala Ser Asn Ser Val Asp
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Glu Thr Asp Pro Thr Gln Asp Asn Trp Gly Tyr Asp Pro Arg Asn Tyr
500 505 510

Asp Val Pro Glu Gly Gln Tyr Ala Thr Asn Ala Asn Gly Asn Ala Arg
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Gly Val Asn Met Asp Val Val Tyr Asn His Thr Phe Ala Thr Gln Ile
545 550 555 560

Ser Asp Phe Asp Lys Ile Val Pro Glu Tyr Tyr Tyr Arg Thr Asp Asp
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Ala Gly Asn Tyr Thr Asn Gly Ser Gly Thr Gly Asn Glu Ile Ala Ala
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Glu Arg Pro Met Val Gln Lys Phe Ile Ile Asp Ser Leu Lys Tyr Trp
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Val Asn Glu Tyr His Ile Asp Gly Phe Arg Phe Asp Leu Met Ala Leu
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Leu Gly Lys Asp Thr Met Ser Lys Ala Ala Ser Glu Leu His Ala Ile
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Gly	Val	Ala 675	Val	Phe	Asn	Asp	Asn 680	Leu	Arg	Asn	Ala	Leu 685	Asp	Gly	Asn
Val	Phe 690	Asp	Ser	Ser	Ala	Gln 695	Gly	Phe	Ala	Thr	Gly 700	Ala	Thr	Gly	Leu
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Asp	Arg	Ile 755	Lys	Met	Asp	Glu	Leu 760	Ala	Gln	Ala	Val	Val 765	Met	Thr	Ser
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Asp	Trp	Ser	Arg	Lys 805	Ala	Gln	Tyr	Pro	Asp 810	Val	Phe	Asn	Tyr	Tyr 815	Ser
Gly	Leu	Ile	His 820	Leu	Arg	Leu	Asp	His 825	Pro	Ala	Phe	Arg	Met 830	Thr	Thr
Ala	Asn	Glu 835	Ile	Asn	Ser	His	Leu 840	Gln	Phe	Leu	Asn	Ser 845	Pro	Glu	Asn
Thr	Val 850	Ala	Tyr	Glu	Leu	Thr 855	Asp	His	Val	Asn	Lys 860	Asp	Lys	Trp	Gly
Asn 865	Ile	Ile	Val	Val	Tyr 870	Asn	Pro	Asn	Lys	Thr 875	Val	Ala	Thr	Ile	Asn 880
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885

890

895

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 Asn Tyr Ala Asn Trp Asp Leu Trp Met Trp Pro Tyr Gln Pro Val Asn
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 Gly Asn Gly Ala Ala Tyr Glu Phe Ser Gly Lys Asp Asp Phe Gly Val
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aaa gca gat gtt caa gtg cct ggg gat gat aca cag gta ggt ctg att 192
 Lys Ala Asp Val Gln Val Pro Gly Asp Asp Thr Gln Val Gly Leu Ile
 50 55 60

gtc cgt aca aat gat tgg agc caa aaa aat aca tca gac gat ctc cat 240
 Val Arg Thr Asn Asp Trp Ser Gln Lys Asn Thr Ser Asp Asp Leu His
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 Ile Asp Leu Thr Lys Gly His Glu Ile Trp Ile Val Gln Gly Asp Pro
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 Asn Ile Tyr Tyr Asn Leu Ser Asp Ala Gln Ala Ala Ala Thr Pro Lys
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aaa Lys	aat Asn 610	aga Arg	gtt Val	atg Met	gga Gly	agt Ser 615	att Ile	tca Ser	gat Asp	ttc Phe	act Thr 620	tcg Ser	gca Ala	cct Pro	agc Ser	1872
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Phe Met Gln 675 Gly Gly Glu Glu Met 680 Leu Arg Thr Lys 685 Gly Gly Asn Asp	
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Asn Ser 690 Tyr Asn Ala Gly Asp 695 Ser Val Asn Gln Phe 700 Asp Trp Ser Arg	
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Lys Ala Gln Phe Glu 710 Asn Val Phe Asp Tyr 715 Ser Trp Leu Ile His 720	
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Leu Arg Asp Asn His 725 Pro Ala Phe Arg Met 730 Thr Thr Ala Asp Gln 735 Ile	
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Lys Gln Asn Leu 740 Thr Phe Leu Asp Ser 745 Pro Thr Asn Thr Val 750 Ala Phe	
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Met Tyr 770 Asn Pro Asn Lys Thr 775 Ala Gln Thr Leu 780 Thr Leu Pro Ser Gly	
aat tgg aca att gta gga tta ggc aat caa gta ggt gag aaa tca cta	2400
Asn Trp Thr Ile Val 790 Gly Leu Gly Asn Gln Val 795 Gly Glu Lys Ser Leu 800	
ggc cat gta aat ggc acg gtt gag gtg cca gct ctt agt acg atc att	2448
Gly His Val Asn 805 Gly Thr Val Glu Val 810 Pro Ala Leu Ser Thr 815 Ile Ile	
ctt cat cag ggt aca tct gaa gat gtc att gat caa aat	2487
Leu His Gln 820 Gly Thr Ser Glu Asp Val 825 Ile Asp Gln Asn	

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 Gly Asn Gly Ala Ala Tyr Glu Phe Ser Gly Lys Asp Asp Phe Gly Val
 35 40 45
 Lys Ala Asp Val Gln Val Pro Gly Asp Asp Thr Gln Val Gly Leu Ile
 50 55 60
 Val Arg Thr Asn Asp Trp Ser Gln Lys Asn Thr Ser Asp Asp Leu His
 65 70 75 80
 Ile Asp Leu Thr Lys Gly His Glu Ile Trp Ile Val Gln Gly Asp Pro
 85 90 95
 Asn Ile Tyr Tyr Asn Leu Ser Asp Ala Gln Ala Ala Ala Thr Pro Lys
 100 105 110
 Val Ser Asn Ala Tyr Leu Asp Asn Glu Lys Thr Val Leu Ala Lys Leu
 115 120 125
 Thr Asn Pro Met Thr Leu Ser Asp Gly Ser Ser Gly Phe Thr Val Thr
 130 135 140
 Asp Lys Thr Thr Gly Glu Gln Ile Pro Val Thr Ala Ala Thr Asn Ala
 145 150 155 160
 Asn Ser Ala Ser Ser Ser Glu Gln Thr Asp Leu Val Gln Leu Thr Leu
 165 170 175
 Ala Ser Ala Pro Asp Val Ser His Thr Ile Gln Val Gly Ala Ala Gly
 180 185 190
 Tyr Glu Ala Val Asn Leu Ile Pro Arg Asn Val Leu Asn Leu Pro Arg
 195 200 205
 Tyr Tyr Tyr Ser Gly Asn Asp Leu Gly Asn Val Tyr Ser Asn Lys Ala
 210 215 220
 Thr Ala Phe Arg Val Trp Ala Pro Thr Ala Ser Asp Val Gln Leu Leu
 225 230 235 240
 Leu Tyr Asn Ser Glu Thr Gly Pro Val Thr Lys Gln Leu Glu Met Gln
 245 250 255
 Lys Ser Asp Asn Gly Thr Trp Lys Leu Lys Val Pro Gly Asn Leu Lys
 260 265 270

Asn Trp Tyr Tyr Leu Tyr Gln Val Thr Val Asn Gly Lys Thr Gln Thr
 275 280 285

Ala Val Asp Pro Tyr Val Arg Ala Ile Ser Val Asn Ala Thr Arg Gly
 290 295 300

Met Ile Val Asp Leu Glu Asp Thr Asn Pro Pro Gly Trp Lys Glu Asp
 305 310 315 320

His Gln Gln Thr Pro Ala Asn Pro Val Asp Glu Val Ile Tyr Glu Val
 325 330 335

His Val Arg Asp Phe Ser Ile Asp Ala Asn Ser Gly Met Lys Asn Lys
 340 345 350

Gly Lys Tyr Leu Ala Phe Thr Glu His Gly Thr Lys Gly Pro Asp Asn
 355 360 365

Val Lys Thr Gly Ile Asp Ser Leu Lys Glu Leu Gly Ile Asn Ala Val
 370 375 380

Gln Leu Gln Pro Ile Glu Glu Phe Asn Ser Ile Asp Glu Thr Gln Pro
 385 390 395 400

Asn Met Tyr Asn Trp Gly Tyr Asp Pro Arg Asn Tyr Asn Val Pro Glu
 405 410 415

Gly Ala Tyr Ala Thr Thr Pro Glu Gly Thr Ala Arg Ile Thr Gln Leu
 420 425 430

Lys Gln Leu Ile Gln Ser Ile His Lys Asp Arg Ile Ala Ile Asn Met
 435 440 445

Asp Val Val Tyr Asn His Thr Phe Asn Val Gly Val Ser Asp Phe Asp
 450 455 460

Lys Ile Val Pro Gln Tyr Tyr Tyr Arg Thr Asp Ser Ala Gly Asn Tyr
 465 470 475 480

Thr Asn Gly Ser Gly Val Gly Asn Glu Ile Ala Thr Glu Arg Pro Met
 485 490 495

Val Gln Lys Phe Val Leu Asp Ser Val Lys Tyr Trp Val Lys Glu Tyr
 500 505 510

His Ile Asp Gly Phe Arg Phe Asp Leu Met Ala Leu Leu Gly Lys Asp
 515 520 525

Thr Met Ala Lys Ile Ser Lys Glu Leu His Ala Ile Asn Pro Gly Ile
530 535 540

Val Leu Tyr Gly Glu Pro Trp Thr Gly Gly Thr Ser Gly Leu Ser Ser
545 550 555 560

Asp Gln Leu Val Thr Lys Gly Gln Gln Lys Gly Leu Gly Ile Gly Val
565 570 575

Phe Asn Asp Asn Ile Arg Asn Gly Leu Asp Gly Asn Val Phe Asp Lys
580 585 590

Ser Ala Gln Gly Phe Ala Thr Gly Asp Pro Asn Gln Val Asn Val Ile
595 600 605

Lys Asn Arg Val Met Gly Ser Ile Ser Asp Phe Thr Ser Ala Pro Ser
610 615 620

Glu Thr Ile Asn Tyr Val Thr Ser His Asp Asn Met Thr Leu Trp Asp
625 630 635 640

Lys Ile Ser Ala Ser Asn Pro Asn Asp Thr Gln Ala Asp Arg Ile Lys
645 650 655

Met Asp Glu Leu Ala Gln Ala Val Val Phe Thr Ser Gln Gly Val Pro
660 665 670

Phe Met Gln Gly Gly Glu Glu Met Leu Arg Thr Lys Gly Gly Asn Asp
675 680 685

Asn Ser Tyr Asn Ala Gly Asp Ser Val Asn Gln Phe Asp Trp Ser Arg
690 695 700

Lys Ala Gln Phe Glu Asn Val Phe Asp Tyr Tyr Ser Trp Leu Ile His
705 710 715 720

Leu Arg Asp Asn His Pro Ala Phe Arg Met Thr Thr Ala Asp Gln Ile
725 730 735

Lys Gln Asn Leu Thr Phe Leu Asp Ser Pro Thr Asn Thr Val Ala Phe
740 745 750

Glu Leu Lys Asn His Ala Asn His Asp Lys Trp Lys Asn Ile Ile Val
755 760 765

Met Tyr Asn Pro Asn Lys Thr Ala Gln Thr Leu Thr Leu Pro Ser Gly
770 775 780

Asn Trp Thr Ile Val Gly Leu Gly Asn Gln Val Gly Glu Lys Ser Leu
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Gly His Val Asn Gly Thr Val Glu Val Pro Ala Leu Ser Thr Ile Ile
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